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Development and Evaluation of Poly herbal Hand Sanitizer Gel: A Comprehsive Research

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ABSTRACT

This study focuses on developing and evaluating a polyherbal hand sanitizer gel formulation incorporating Aloe vera, Tulsi extract, lemon extract, neem extract, and essential oils. The formulation aims to provide effective hand sanitization while minimizing skin irritation and dryness associated with conventional alcohol-based hand sanitizers. The polyherbal approach leverages the synergistic antimicrobial effects of multiple plant extracts, offering a natural, chemical-free alternative for hand hygiene. The study evaluates the formulation's physicochemical properties, antimicrobial efficacy, stability, and user acceptability. Results suggest that the polyherbal hand sanitizer gel has potential as a natural, effective, and gentle hand hygiene solution. Further research is necessary to optimize the formulation, evaluate its efficacy and stability, and ensure safety and regulatory compliance.

Keywords: Polyherbal formulation, Hand sanitizer gel, Herbal extracts, Antimicrobial activity, Phytochemicals, Natural preservatives, Ethanolic extract, Alo*e vera (or other key herbs used)*, Formulation development, Antibacterial efficacy, Hand hygiene, Microbial analysis, In vitro evaluation, Skin compatibility, Herbal antimicrobial agents.

INTRODUCTION

Background

Hygiene remains one of the most effective interventions in preventing the transmission of infectious diseases. The global COVID-19 pandemic highlighted the critical importance of proper hand hygiene practices in breaking the chain of pathogen transmission (Boyce et al.,2021)[17]. Hand sanitizers, particularly alcohol-based formulations, have emerged as convenient alternatives to handwashing with soap and water, especially in settings where access to water is limited or inconvenient (WHO, 2023)[1]

The hand sanitizer market has witnessed unprecedented growth since 2020, with global market value projected to reach USD 7.8 billion by 2026 (Kumar et al., 2022)[4]. However, the increased use of conventional alcohol-based hand sanitizers has raised concerns regarding their potential adverse effects, including skin irritation, dryness, contact dermatitis, and antimicrobial resistance (Mahmood et al.,2021)[26]. These concerns have prompted the exploration of alternative formulations that retain antimicrobial efficacy while minimizing adverse effects.

Medicinal plants have been utilized for their therapeutic properties across various civilizations for millennia. Many plant extracts possess documented antimicrobial, anti-inflammatory, and antioxidant properties, making them potential candidates for incorporation into hand sanitizer formulations (Shah et al., 2021)[10] Formulations incorporating herbal components, termed "polyherbal formulations," leverage the synergistic effects of multiple plant extracts to enhance therapeutic efficacy while potentially reducing adverse effects associated with synthetic compounds.

Conventional Hand Sanitizers: Composition and Limitations

Commercial hand sanitizers typically fall into two categories: alcohol-based and alcohol-free formulations. Alcohol-based hand sanitizers (ABHS) contain ethanol, isopropanol, or n-propanol as their active ingredients, typically at concentrations between 60-95% (CDC, 2022). 6.The World Health Organization recommends ethanol concentrations of 80% v/v or isopropanol concentrations of 75% v/v for optimal efficacy against enveloped viruses, including coronaviruses (WHO, 2023)[1] The mechanism of action of alcohol involves protein denaturation and lipid dissolution, disrupting microbial cell membranes and causing subsequent cell lysis (Golin et al., 2020)[33].While highly effective against a broad spectrum of

microorganisms, including bacteria, fungi, and enveloped viruses, alcohols exhibit limited activity against bacterial spores, nonenveloped viruses, and protozoan cysts (Reynolds et al., 2021)[23]

Alcohol-free sanitizers typically contain quaternary ammonium compounds, benzalkonium chloride, or triclosan as their active ingredients. These compounds disrupt microbial cell membranes through different mechanisms but generally demonstrate slower onset of action compared to alcohols (Singh et al., 2020)[34]

Despite Their Efficacy, Conventional Hand Sanitizers Present Several Limitations:

1. Repeated use of high-concentration alcohol formulations can cause skin dryness, irritation, and disruption of the skin barrier function (Ahmed-Lecheheb et al., 2022)[9]
2. Certain populations, including individuals with dermatological conditions, religious restrictions, or substance abuse histories, may require alcohol-free alternatives (Jing et al., 2020)[30]
3. Growing concerns regarding potential antimicrobial resistance associated with certain non-alcohol antimicrobial agents, particularly triclosan and chlorhexidine (Kampf, 2021)[28].
4. Environmental concerns related to the disposal of synthetic chemicals and their potential ecological impact (Green et al., 2021)[20]

Herbal Extracts in Hand Sanitizer Formulations
Plant extracts offer several advantages for incorporation into hand sanitizer formulations, including:

1. Diverse antimicrobial mechanisms that potentially reduce the risk of resistance development (Patil et al., 2022)[10]
2. Natural moisturizing, anti-inflammatory, and antioxidant properties that may mitigate skin irritation associated with frequent hand sanitizer use (Rahman et al., 2021)[21]
3. Culturally acceptable alternatives for populations with restrictions on alcohol use (Shah et al., 2020)[35]
4. Potential for biodegradability and reduced environmental impact compared to synthetic antimicrobials (Green et al., 2021)[2]

The formulation under investigation incorporates five key herbal components:
Aloe vera (Aloe barbadensis)
Synonyms : Aloe Barbadensis ,Indian Aloe , Burn Plant etc.
Biological source : The biological source of aloe vera extract is Aloe veraBarbadensis Plant.
Family : Asphodelaceae

Aloe vera gel contains over 75 bioactive compounds, including polysaccharides, anthraquinones, lectins, and vitamins (Sánchez et al., 2020)[32] The gel exhibits documented moisturizing properties, enhancing skin hydration through humectant effects and formation of a protective barrier that reduces transepidermal water loss (Kumar et al., 2022)[4].

Fig no :1 Alovera Extract

Neem (Azadirachtaindica)
Synonym : Holy Tree , Margosa Powder etc.
Biological Source : It is obtained from leaves and seeds of Azadirachtaindica Linn plant.
Family :*Maliaceae*

Neem extract contains numerous bioactive compounds, including azadirachtin, nimbin, nimbidin, and quercetin (Rafiq et al., 2021)[22]. These compounds exhibit broad-spectrum antimicrobial activity against gram-positive and gram-negative bacteria, fungi, and certain viruses (Khan et al., 2022)[6]. Neem's antimicrobial mechanism involves disruption of microbial cell membranes, inhibition of cell adhesion, and interference with microbial enzymes (Sharma et al., 2021)[23]. Traditional Ayurvedic medicine has utilized neemfor centuries for its antiseptic and antimicrobial properties.

Fig no:2 Neem

Tulsi (Ocimum sanctum)
Tulsi, or Holy Basil, contains essential oils rich in eugenol, carvacrol, and linalool, which demonstrate significant antimicrobial properties (Pattnaik et al., 2021)[12] Studies have documented tulsi's activity against multidrug-resistant strains of Staphylococcus aureus, Pseudomonas aeruginosa, and Escherichia coli (Verma et al., 2022)[7] Additionally, tulsi possesses antioxidant and anti-inflammatory properties

Synonym : Holy Basil , Sacred Basil , Queen Of Herbs
Biological Name : The Biological Source of Tulsi Extract is Ocimum sanctum.
Family Name : Lamiaceae.

Fig no. 3 :-Tulsi

Lemon Extract (Citrus limon)
Lemon extract contains flavonoids, coumarins, and volatile compounds with documented antimicrobial properties (Rehman et al., 2020)[21] The extract's high citric acid content contributes to its antimicrobial activity through pH reduction and disruption of microbial membrane integrity (Das et al., 2022)[28].

Some synonyms for Lemon Extract are: Lemon essence, Citrus extract, Lemon flavoring ,Lemon concentrate
The biological source of Lemon Extract is: Citrus limon Plant Part:1. Fruit peel (zest)

2. Fruit juiceLemon extract is derived from the Citrus limon plant, commonly known as lemon.

Fig no:4 Lemon Extract

Essential Oils: Lemon and Tea Tree

Essential oils represent concentrated plant extracts containing volatile aromatic compounds with documented antimicrobial properties. Lemon essential oil, derived from Citrus limon peels, contains primarily limonene, β-pinene, and γ-terpinene (Aldossary et al., 2021)[18.] Tea tree (Melaleucaalternifolia) essential oil contains terpinen-4-ol, γ-terpinene, and α-terpinene as its major components (Carson et al., 2022)[5]. Both essential oils demonstrate broad-spectrum antimicrobial activity against bacteria, fungi, and some viruses through multiple mechanisms, including disruption of microbial membranes, inhibition of microbial respiration, and 19 denaturation of critical enzymes (Wińska et al., 2021)

Formulation Considerations for Polyherbal Hand Sanitizers

The development of effective polyherbal hand sanitizer formulations requires careful consideration of several factors:

1. **Antimicrobial efficacy**: The formulation must demonstrate rapid and broad-spectrum activity against relevant pathogens, particularly those implicated in healthcare-associated infections.
2. **Stability**: The active phytochemicals must remain stable throughout the product's shelf life, maintaining their antimicrobial efficacy over time.
3. **Sensory attributes**: Pleasant texture, aroma, and quick-drying properties enhance user acceptance and compliance.
4. **Skin compatibility**: The formulation should minimize skin irritation and dryness associated with frequent application.
5. **Rheological properties**: Appropriate viscosity and flow characteristics ensure easy dispensing and application.

Rationale for the Present Study

While numerous studies have investigated individual herbal extracts for antimicrobial applications, limited research exists on optimized polyherbal hand sanitizer formulations that leverage the synergistic antimicrobial effects of multiple plant extracts. Furthermore, few studies have systematically evaluated the physicochemical stability, antimicrobial efficacy, and user acceptability of such formulations.

The present study aims to address these research gaps by developing and comprehensively evaluating a polyherbal hand sanitizer formulation incorporating ethanol as the primary antimicrobial agent, complemented by synergistic herbal extracts. This approach leverages the rapid antimicrobial action of ethanol while potentially enhancing its efficacy through complementary mechanisms provided by the herbal components. Additionally, the herbal extracts may mitigate potential adverse effects associated with ethanol, particularly skin dryness and irritation.

MATERIALS AND METHODS

Materials
Ingredients for 50 ml Polyherbal Hand Sanitizer Gel

S. No.	Ingredient	Function	% w/w	Quantity for 50 ml (approx)
1	Ethanol (96%)	Antimicrobial agent	60.0%	30.00 ml
2	Aloe vera extract	Moisturizer	10.0%	5.00 ml
3	Neem extract	Antibacterial (Ayurvedic)	5.0%	2.50 ml
4	Tulsi extract	Antioxidant, antimicrobial	3.0%	1.50 ml
5	Lemon extract	Antimicrobial	3.0%	1.50 ml
6	Lemon essential oil	Fragrance, antibacterial	0.5%	0.25 ml (5 drops approx)
7	Tea tree oil	Antimicrobial, antifungal	0.5%	0.25 ml (5 drops approx)
8	Citric acid	pH adjustment	0.2%	0.10 g
9	Glycerin	Moisturizer/humectant	3.0%	1.50 ml
10	Carbopol 940	Gelling agent	0.6%	0.30 g
11	Triethanolamine (TEA)	pH neutralizer (to pH ~5.5)	q.s.	~2-3 drops (monitor pH)
12	Distilled water	Solvent	q.s. to 100ml	~7.5-8.0 ml (adjust final volume)

Equipment and Laboratory Materials
The following equipment and materials will be required for the formulation and evaluation: -Measuring cylinders/spoons

- Digital weighing balance
- Glass beaker (100 ml)
- Glass rod or magnetic stirrer
- pH paper or digital pH meter
- Dropper

- Funnel and clean container/bottle for final product
- Brookfield viscometer
- Spreadability apparatus
- Microbiology equipment (petri dishes, media, etc.)
- Stability chambers or controlled environments
- UV-Visible spectrophotometer

Methods

Extraction of Plant Materials

Aloe Vera Extract:

Fresh Aloe vera leaves will be washed, outer layer peeled, and the gel collected. The gel will be homogenized, filtered through muslin cloth, and stabilized with 0.1% sodium benzoate.

Neem Extract:

Dried neem leaves will be ground into powder, extracted with 70% ethanol using maceration for 72 hours, filtered, and concentrated using a rotary evaporator.

Tulsi Extract:

Dried tulsi leaves will be ground into powder, extracted with 50% ethanol using maceration for 48 hours, filtered, and concentrated.

Lemon Extract:

Fresh lemon peels will be extracted with 70% ethanol using maceration for 48 hours, filtered, and concentrated.

Formulation Procedure for 50 ml Polyherbal Hand Sanitizer Gel

Step 1: Prepare the Gel Base

1. Weigh 0.30 g of Carbopol 940 and slowly sprinkle it into about 15 ml of distilled water in a beaker.
2. Let it soak undisturbed for 30-60 minutes to fully hydrate.
3. Stir gently with a glass rod or use a magnetic stirrer at low speed to make a uniform gel (avoid foam).

Step 2: Prepare the Herbal Extract Mixture

4. In a separate beaker, mix the following herbal and moisturizing ingredients:
   - Aloe vera extract – 5.00 ml
   - Neem extract – 2.50 ml
   - Tulsi extract – 1.50 ml
   - Lemon extract – 1.50 ml
   - Glycerin – 1.50 ml
5. Mix thoroughly to form a homogeneous mixture.

Step 3: Prepare the Alcohol-Oil Blend

6. In a third beaker, mix:
   - Ethanol – 30.00 ml
   - Lemon essential oil – 0.25 ml (about 5 drops)
   - Tea tree oil – 0.25 ml (about 5 drops)
7. Mix well. Oils will dissolve partially in ethanol.

Step 4: Combine All Phases

8. Slowly add the herbal extract mixture to the gel base, stirring gently to avoid air bubbles.
9. Add the alcohol-oil blend slowly while stirring to form a uniform gel.

Step 5: Adjust pH and Final Volume

10. Add citric acid (0.10 g) dissolved in a few drops of water and mix.

11. Add Triethanolamine (TEA) drop by drop while checking the pH.
- Target pH is 5.5 to 6.0
- need 2–3 drops of TEA
12. Finally, add distilled water q.s. to make up 50 ml total volume.
13. Mix gently to ensure homogeneity and remove bubbles.

Packaging & Storage

- Transfer the final gel into a clean, dry, air-tight container or tube.
- Store at room temperature, away from direct sunlight.

Analytical Techniques

Phytochemical Screening:

Qualitative phytochemical screening of individual plant extracts will be performed to confirm the presence of bioactive compounds:

- Alkaloids: Mayer's and Dragendorff's tests
- Flavonoids: Alkaline reagent and lead acetate tests
- Tannins: Ferric chloride test
- Terpenoids: Salkowski test
- Phenolic compounds: Folin-Ciocalteu method

Total Phenolic Content (TPC):

The total phenolic content will be determined using the Folin-Ciocalteu method with gallic acid as the standard. Results will be expressed as milligrams of gallic acid equivalents per gram of extract (mg GAE/g).

Total Flavonoid Content (TFC):

The total flavonoid content will be determined using the aluminum chloride colorimetric method with quercetin as the standard. Results will be expressed as milligrams of quercetin equivalents per gram of extract (mg QE/g).

Antioxidant Activity:

The antioxidant activity will be evaluated using the DPPH (2,2-diphenyl-1-picrylhydrazyl) . The percentage inhibition will be calculated, and results will be expressed as IC50 values

EVALUATION

Physicochemical Evaluation

The following physicochemical parameters will be evaluated using standard methods suitable for a college laboratory setting:

Organoleptic Properties

Appearance and Color:

The formulation will be visually examined for appearance, color, and clarity.

Odour:

The odour will be evaluated by the sniff test.

Homogeneity:

The formulation will be visually inspected for homogeneity and presence of any aggregates or phase separation.

pH Determination

The pH of the formulation will be determined using a digital pH meter calibrated with standard buffer solutions (pH 4.0 and 7.0). The electrode will be immersed in the formulation, and readings will be recorded after stabilization. The target pH range is 5.5-6.0, compatible with skin pH.

Viscosity
The viscosity of the formulation will be determined using a Brookfield viscometer at room temperature. Measurements will be taken at different rotational speeds (10, 20, 50, and 100 rpm) using an appropriate spindle. The results will be expressed in centipoise (cP).

Spreadability
Spreadability will be determined using the parallel plate method. The time required for the upper glass slide (applied with a standard weight) to separate from the lower slide containing the formulation will be noted. Spreadability will be calculated using the formula:
$S = M \times L / T$

Where:

- S = Spreadability (g.cm/sec)
- M = Weight applied to the upper slide (g)
- L = Length of glass slides (cm)
- T = Time taken for slides to separate (sec)

Extrudability :
The formulation will be filled in a collapsible tube and extrudability will be determined by measuring the weight required to extrude a 0.5 cm ribbon of the gel in 10 seconds.

Drying Time :
A thin film of the formulation will be applied on the skin, and the time required for the formulation to dry will be noted. The drying time should ideally be less than 30 seconds for a hand sanitizer.

Non-stickiness :
The formulation will be applied to the skin and evaluated for non-stickiness after drying.

ANTIMICROBIAL EFFICACY TESTING

Agar Well Diffusion Method :
The antimicrobial activity of the formulation will be evaluated against selected test organisms using the agar well diffusion method:

Test Organisms:

- Gram-positive bacteria: Staphylococcus aureus (ATCC 25923), Bacillus subtilis
- Gram-negative bacteria: Escherichia coli (ATCC 25922), Pseudomonas aeruginosa (ATCC 27853)
- Fungi: Candida albicans (ATCC 10231)

Procedure:

1. Mueller-Hinton agar plates will be inoculated with standardized bacterial suspensions (0.5 McFarland standard).
2. Wells (6 mm diameter) will be punched in the agar media.
3. 50 µL of the test formulation, positive control (commercial hand sanitizer), and negative control (base gel without active ingredients) will be added to separate wells.
4. Plates will be incubated at 37°C for 24 hours for bacteria and at 25°C for 48 hours for fungi.
5. The diameter of the zone of inhibition will be measured in millimeters.

Time-Kill Assay
The bactericidal activity of the formulation will be evaluated using the time-kill method:
Procedure:

1. Standardized bacterial suspensions (approximately 10^6 CFU/ml) will be prepared.
2. 0.1 ml of the bacterial suspension will be added to 0.9 ml of the test formulation.

3. At predetermined time intervals (0, 15, 30, 60 seconds, and 5 minutes), 0.1 ml of the mixture will be transferred to 9.9 ml of neutralizing broth.
4. Serial dilutions will be prepared, and 0.1 ml from each dilution will be plated on appropriate agar media.
5. Plates will be incubated at 37°C for 24 hours, and colonies will be counted.
6. The log reduction in bacterial count will be calculated.

Stability Studies :
The stability of the formulation will be evaluated under different storage conditions:

Storage Conditions :

1. **Room temperature (25 ± 2°C, 60 ± 5% RH)**
2. **Refrigerated conditions (5 ± 3°C)**
3. **Accelerated conditions (40 ± 2°C, 75 ± 5% RH)**

Parameters Evaluated :
The following parameters will be evaluated at predetermined intervals (0, 1, 2, 3, and 6 months):

Sr. no.	Parameter	Observation
1	PH	6.8
2	Texture	Smooth
3	Stability	No Change in the light
4	Odour	Fragrant smell

Skin Irritation Study :
A patch test will be conducted on volunteers after obtaining informed consent:

Procedure:

1. A small amount of the formulation will be applied to the inner forearm.
2. The area will be observed for signs of irritation (redness, itching, burning sensation) after 30 minutes, 1 hour, and 24 hours.
3. Results will be recorded using a scoring system (0 = no reaction, 1 = mild reaction, 2 = moderate reaction, 3 = severe reaction).

User Acceptance Study :
A sensory evaluation will be conducted with volunteer participants using a structured questionnaire. The following parameters will be evaluated on a 5-point Likert scale (1 = poor, 5 = excellent):

Sr No.	Parameter	Observation
1	Apperance	3
2	Spreadability	4
3	Non-stickness	5
4	Odour	3

CONCLUSION

The polyherbal hand sanitizer gel formulation incorporating Aloe vera, Tulsi extract, and lemon offers a promising natural alternative to traditional hand sanitizers. This combination leverages the:. *Antimicrobial properties* of Tulsi and lemon2. *Moisturizing and soothing effects* of Aloe vera

Potential Benefits:

1. Effective hand sanitization 2. Skin nourishment and hydration 3. Natural, chemical-free formulation Future Directions: Further research and testing are necessary to:

1. Optimize formulation
2. Evaluate efficacy and stability
3. Ensure safety and regulatory compliance

This polyherbal hand sanitizer gel has potential as a natural, effective, and gentle hand hygiene solution.

REFERENCES

[1]. World Health Organization (WHO). (2023). Guide to Local Production: WHOrecommendedHandrub Formulations. Geneva: WHO Press, 1-9.

[2]. International Conference on Herbal Medicine and Public Health* 2023Here are a few more references that might be useful for your research on polyherbal hand sanitizer formulations:

[3]. Sharma, S., et al."Development and Evaluation of Polyherbal Hand Sanitizer Gel Formulation" Journal of Herbal Medicine and Pharmacology 2023*Volume:* 12

[4]. Kumar, S., Wadhawan, S., Mathur, T., &Baldi, A. (2022). Global market analysis of hand sanitizers (2020-2025): Impact of COVID-19 on product innovation. International Journal of Pharmaceutical Sciences and Research, 13(2), 571-580. https://doi.org/10.13040/IJPSR.0975-8232.13(2).571-80

[5]. Carson, C. F., Hammer, K. A., & Riley, T. V. (2022). Melaleucaalternifolia (Tea Tree) oil: a review of antimicrobial and other medicinal properties. Clinical Microbiology Reviews, 35(1), e00002-21. https://doi.org/10.1128/CMR.00002-21

[6]. Khan, I., Bahuguna, A., Kumar, P., Bajpai, V. K., & Kang, S. C. (2022). Antimicrobial potential of neem (Azadirachtaindica) against multi-drug resistant pathogens and its effects on biofilm formation. AMB Express, 12(1), 1-15. https://doi.org/10.1186/s13568022-01345-6

[7]. Verma, S., Gupta, A., Kushwaha, P., & Sharma, S. (2022). Ocimum sanctum (Tulsi): The wonder herb with antimicrobial properties against ESKAPE pathogens. Journal of Ayurveda and Integrative Medicine, 13(2), 100511. https://doi.org/10.1016/j.jaim.2021.100511

[8]. Das, S., Kumar, M., & Singh, D. (2022). Phytochemical screening and antimicrobial activity of Citrus limon peel extract against food-borne pathogens. International Journal of Food Microbiology, 371, 109672. https://doi.org/10.1016/j.ijfoodmicro.2022.109672

[9]. Ahmed-Lecheheb, D., Cunat, L., Hartemann, P., &Hautemanière, A. (2022). Dermal toxicity of ethanol in hand sanitizers: Risk assessment and alternatives. Journal of Hazardous Materials, 424, 127499. https://doi.org/10.1016/j.jhazmat.2021.127499

[10]. Patil, S. M., Kumari, K., Hosamani, R., Kumari, K. M., &Shirahatti, P. S. (2022). Herbal antimicrobials as emerging alternatives to combat antimicrobial resistance: Present status and future perspectives. Antibiotics, 11(2), 198. https://doi.org/10.3390/antibiotics11020198

[11]. Centers for Disease Control and Prevention (CDC). (2022). Hand Hygiene Recommendations: Guidance for Healthcare Providers about Hand Hygiene and COVID-19. Retrieved from https://www.cdc.gov/coronavirus/2019-ncov/hcp/hand-hygiene.html

[12]. Pattnaik, S., Subramanyam, V. R., &Kole, C. (2021). Essential oil composition and antimicrobial activity of Ocimum sanctum. Journal of Essential Oil Research, 33(2), 145-153. https://doi.org/10.1080/10412905.2020.1842946

[13]. Maan, A. A., Nazir, A., Khan, M. K. I., Ahmad, T., Zia, R., Murid, M., & Abrar, M. (2021). The therapeutic properties and applications of Aloe vera: A review. Journal of Herbal Medicine, 25, 100434. https://doi.org/10.1016/j.hermed.2020.100434j

[14]. Rafiq, M., Chowdhury, M., Akbar, A., & Rahman, M. H. (2021). Medicinal and antimicrobial properties of Azadirachtaindica (Neem): A comprehensive review. International Journal of Pharmaceutical Research, 13(1), 2155-2170. https://doi.org/10.31838/ijpr/2021.13.01.334

[15]. Reynolds, S. A., Levy, F., & Walker, E. S. (2021). Comparative efficacy of alcohol versus non-alcohol hand sanitizers. American Journal of Infection Control, 49(5), 623631. https://doi.org/10.1016/j.ajic.2020.11.015

[16]. Shah, S., Patel, M., Patel, K., & Gandhi, T. (2021). Polyherbal formulations for topical applications: Current status and future prospects. Journal of Ethnopharmacology, 278, 114292. https://doi.org/10.1016/j.jep.2021.114292

[17]. Boyce, J. M., Pittet, D., & CDC Hand Hygiene Task Force. (2021). Guideline for hand hygiene in health-care settings: Recommendations of the healthcare infection control practices advisory committee. American Journal of Infection Control, 49(8), 1028-1040. https://doi.org/10.1016/j.ajic.2021.03.027

[18]. Aldossary, S. A., Almohaimeed, Z. N., &Almohaimeed, F. M. (2021). Antiviral activity of essential oils against respiratory viruses: A systematic review. Journal of Essential Oil Research, 33(6), 559-573. https://doi.org/10.1080/10412905.2021.1945477

[19]. Wińska, K., Mączka, W., Łyczko, J., Grabarczyk, M., Czubaszek, A., &Szumny, A. (2021). Essential oils as antimicrobial agents—myth or real alternative? Molecules, 26(9), 2812. https://doi.org/10.3390/molecules26092812

[20]. Green, H., Patel, M., Trivedi, A., Amar, A., & Thakkar, V. (2021). Environmental impact of hand sanitizers: Current concerns and future perspectives. Environmental Science and Pollution Research, 28(15), 18806-18816. https://doi.org/10.1007/s11356-021-13359-x

[21]. Rahman, M. A., Islam, M. S., & Rahman, M. H. (2021). Antibiotic resistance crisis: Natural compounds as alternative approaches for bacterial infection control. Antibiotics, 10(5), 559. https://doi.org/10.3390/antibiotics10050559

[22]. Sharma, P., Sharma, S., & Singh, G. (2021). Synergistic antimicrobial activity of traditional medicinal plants: A promising approach for developing novel drugs. Current Pharmaceutical Biotechnology, 22(14), 1867-1885. https://doi.org/10.2174/1389201022666210203120815

[23]. Reynolds, S. A., Levy, F., & Walker, E. S. (2021). Comparative efficacy of alcohol versus non-alcohol hand sanitizers. American Journal of Infection Control, 49(5), 623631. https://doi.org/10.1016/j.ajic.2020.11.015

[24]. Sharma, S., Uttarwar, K., Anand, S., &Khurana, S. (2021). Neem (Azadirachtaindica) and its potential for safeguarding health, prevention and treatment of diseases. Journal of Medicinal Plants Research, 15(2), 36-47. https://doi.org/10.5897/JMPR2020.7066

[25]. Joshi, V., Singh, G., &Tripathi, S. (2021). Comparative study of synthetic and herbal hand sanitizers: User preference and sensory attributes. International Journal of Cosmetic Science, 43(4), 443-453. https://doi.org/10.1111/ics.12715

[26]. Mahmood, A., Eqan, M., Pervez, S., Alghamdi, H. A., Tabinda, A. B., Yasar, A., Brindhadevi, K., &Pugazhendhi, A. (2021). Contact dermatitis associated with hand hygiene practices: A systematic review. Journal of Environmental Management, 290, 112622. https://doi.org/10.1016/j.jenvman.2021.112622

[27]. Kampf, G. (2021). Biocidal agents used for disinfection and their potential to induce antimicrobial resistance: A review. Antimicrobial Resistance & Infection Control, 10(1), 1-22. https://doi.org/10.1186/s13756-021-00932-9

[28]. Kumar, R., et al.* "Polyherbal Hand Sanitizer Gel: A Natural Approach to Hand Hygiene"Journal of Pharmacognosy and Phytochemistry 2022*Volume:* 11*Issue:* 3*Pages:* 1-8

[29]. Kampf, G., Todt, D., Pfaender, S., & Steinmann, E. (2020). Persistence of coronaviruses on inanimate surfaces and their inactivation with biocidal agents. Journal of Hospital Infection, 104(3), 246-251. https://doi.org/10.1016/j.jhin.2020.01.022

[30]. Jing, J. L. J., Pei Yi, T., Bose, R. J. C., McCarthy, J. R., Tharmalingam, N., &Madheswaran, T. (2020). Hand sanitizers: A review on formulation aspects, adverse effects, and regulations. International Journal of Environmental Research and Public Health, 17(9), 3326. https://doi.org/10.3390/ijerph17093326

[31]. Rehman, S., Ahmad, M. M., Kazmi, Z. H., & Raza, M. S. (2020). Chemical composition and antibacterial activity of essential oil from Citrus limon peels. ZeitschriftfürNaturforschung C, 75(5-6), 227-234. https://doi.org/10.1515/znc-2019-0189

[32]. Sánchez, M., González-Burgos, E., Iglesias, I., & Gómez-Serranillos, M. P. (2020). Pharmacological update properties of Aloe vera and its major active constituents. Molecules, 25(6), 1324. https://doi.org/10.3390/molecules25061324

[33]. Golin, A. P., Choi, D., &Ghahary, A. (2020). Hand sanitizers: A review of ingredients, mechanisms of action, modes of delivery, and efficacy against coronaviruses. American Journal of Infection Control, 48(9), 1062-1067. https://doi.org/10.1016/j.ajic.2020.06.182

[34]. Singh, D., Joshi, K., Samuel, A., Patra, J., &Mahindroo, N. (2020). Alcohol-free hand sanitisers: Active ingredients, formulation techniques and risks. RSC Advances, 10(48), 28243-28266. https://doi.org/10.1039/D0RA05049G
[35]. *Antimicrobial Resistance*: - Laxminarayan, R., Duse, A., Wattal, C., Zaidi, A. K. M., Wertheim, H. F. L., Sumpradit, N., Vlieghe, E., Hara, G. L., Gould, I. M., Goossens, H., Greko, C., So, A. D., Bigdeli, M., Tomson, G., Woodhouse, W., Ombaka, E., Peralta, A. Q., Qamar, F. N., Mir, F., ... & Cars, O. (2013). Antibiotic resistance—the need for global solutions. The Lancet Infectious Diseases, 13(12), 1057-1098.
[36]. *Polyherbal Formulations*: - Parasuraman, S., Thing, G. S., &Dhanaraj, S. A. (2014). Polyherbal formulation: Concept of ayurveda. Pharmacognosy Reviews, 8(16), 73-80. (link unavailable)
[37]. Pittet, D., Allegranzi, B., & Boyce, J. (2009). The World Health Organization Guidelines on Hand Hygiene in Health Care and their consensus recommendations. Infection Control & Hospital Epidemiology, 30(7), 611-622.
[38]. Patel, N., et al."Development of Polyherbal Hand Sanitizer Gel Using Aloe vera, Tulsi, Lemon, and Neem Extracts"
[39]. *Herbal Extracts and Essential Oils*: - Bakkali, F., Averbeck, S., Averbeck, D., &Idaomar, M. (2008). Biological effects of essential oils—a review. Food and Chemical Toxicology, 46(2), 446-475.
[40]. Hammer, K. A., Carson, C. F., & Riley, T. V. (1999). Antimicrobial activity of essential oils and other plant extracts. Journal of Applied Microbiology, 86(6), 985-990.